

**AUSTIN POLICE DEPARTMENT
SEROLOGY/ DNA SECTION
STANDARD OPERATING PROCEDURES**

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Chapter 1 Scope of Operations

1.1 DNA section Overview

The DNA section will follow the guidelines set forth in the Forensic Science Division SOP. Supplemental requirements specific to the DNA section are contained within the Serology/DNA Standard Operating Procedures Manual (SOP), the DNA Technical Manual, and the DNA Training Manual. These manuals combined represent guidelines for the Quality System within the DNA section.

It is not possible to anticipate every situation that may arise or to prescribe a specific course of action for every case; therefore, the examiner must exercise good judgment based on experience and common sense, especially when processing evidence. In some cases, the manual offers guidelines for analysis that must be tempered with the experience of the examiner. However, any portion of a procedure not explicitly qualified as a guideline, e.g., by use of the word “should,” may not be modified for use in casework without prior approval by the Technical Leader.

1.2 History

No Supplemental Requirements

1.3 Mission Statement

No Supplemental Requirements

1.4 Goals and Objectives

No Supplemental Requirements

1.5 Code of Ethics

No Supplemental Requirements

1.6 Organization and Staffing

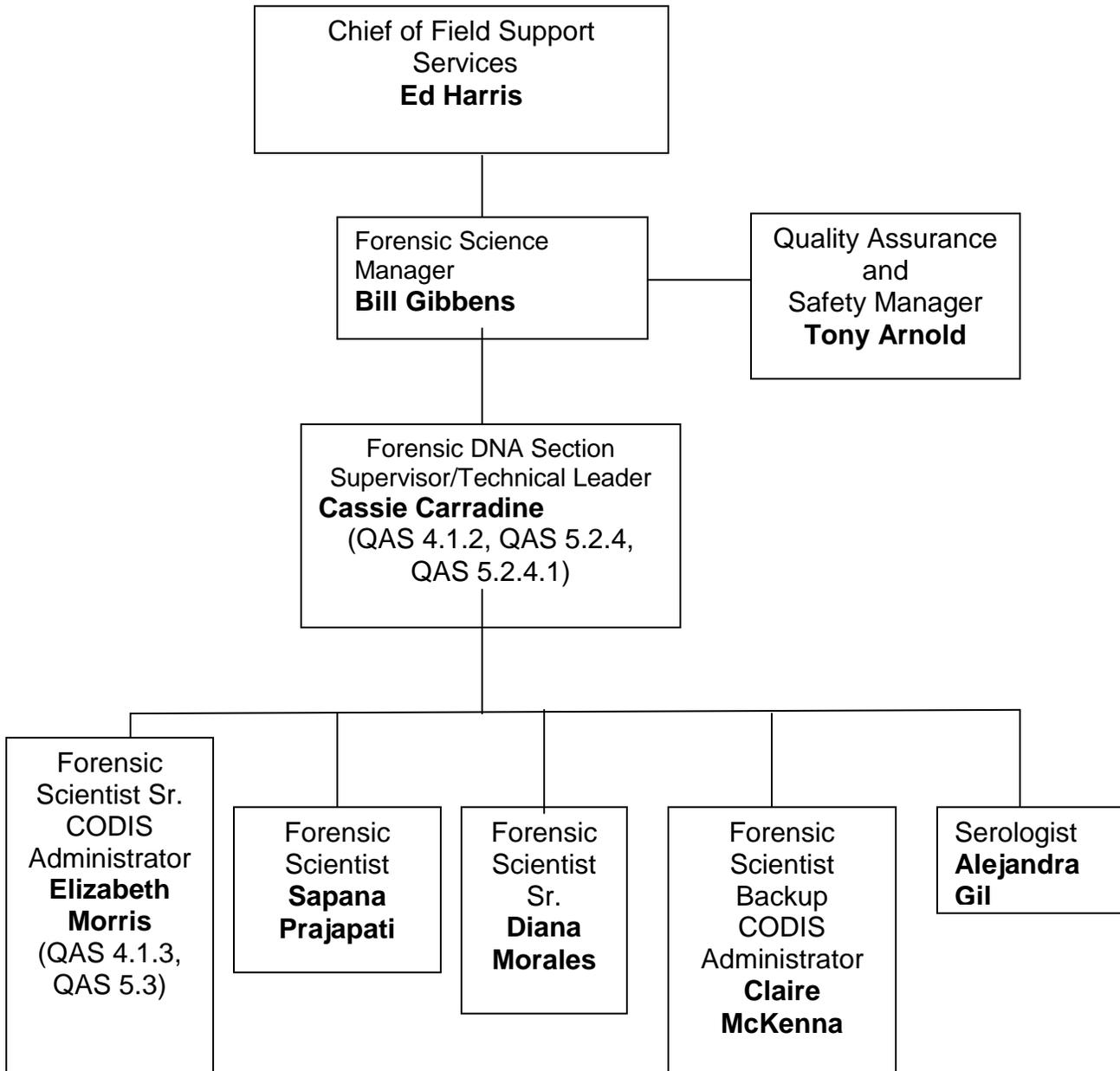
No Supplemental Requirements

1.7 List of Locations, Addresses, and Phone numbers

No Supplemental Requirements

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1.8 Organizational Chart (QAS 3.1.1.2, QAS 4.1.1, QAS 4.1.2, QAS 4.1.3, QAS 4.1.4, QAS 4.1.5, QAS 5.4)



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1.9 Section Descriptions and Responsibilities

No Supplemental Requirements

1.10 Hours of Operation

No Supplemental Requirements

1.11 Manuals

No Supplemental Requirements

1.12 Customer Service

No Supplemental Requirements

1.13 Management System

Authority/Responsibility for the Management System (QAS 3.1.1.3, QAS 4.1.5, QAS 5.1.1)

- DNA Forensic Scientist Supervisor

The DNA Supervisor will have a B.A./B.S. or graduate degree or its equivalent in a biology, chemistry, or forensic science related area. The DNA Supervisor will also have completed college course work in the areas of Biochemistry, Genetics, and Molecular Biology. College course work or training in the subject area of statistics and/or population genetics is also required. If the Supervisor is a working analyst, he/she will be trained in the appropriate procedures and will be current in proficiency testing.

- DNA Technical Leader

The FBI has mandated that each DNA laboratory employ a Technical Leader. The Technical Leader and Supervisor roles may be combined. The Technical Leader is responsible for implementation and execution of valid analysis procedures as well as oversight of training, quality assurance, safety, and proficiency testing.

The Technical Leader shall be accessible to the laboratory during times of laboratory operation.

The Technical Leader is accountable for the laboratory's quality assurance program to the extent that he/she has the authority to terminate the laboratory's or an individual's testing in the event of a technical problem until the problem is resolved (QAS 5.2.3.1.2). Technical Leaders have duties as specified by the FBI for technical leaders in general.

In the event that the DNA Technical Leader is no longer able to perform the duties of Technical Leader, the following plan will immediately take effect (QAS 4.1.6, QAS 5.2.4.1.1):

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- A currently employed and active duty DNA analyst meeting the minimum requirements of the Technical Leader position will be appointed as Interim Technical Leader. The position will then be posted and filled according to City policy within 90 days.
- In the case that no currently employed or active duty analysts meet the minimum requirements of the Technical Leader position, the FBI will be contacted and this contingency plan will be submitted within 14 days. The plan will be that a Technical Leader will be contracted by the laboratory for a maximum of 90 days to allow the City posting and hiring process to take place. During the time that the Technical Leader position becomes vacated and the contingency plan is approved no new DNA casework will be started.

The DNA Technical Leader will have a graduate degree in a biology, chemistry, or forensic science related area and a minimum of 12 credit hours or its equivalent including a combination of graduate and undergraduate coursework covering the subject area of Biochemistry, Genetics, Molecular Biology, and Statistics and/or Population Genetics. If the DNA Technical Leader is a working analyst, he/she will be trained in the appropriate procedures and will be current in proficiency testing.

The Technical Leader is responsible for overseeing the technical operations of the DNA lab and technical problem solving of analytical methods (QAS 5.2.3.1.1). Any technical problem in the laboratory requiring correction beyond routine maintenance by the examiner must be brought to the attention of the Technical Leader.

The duties of the DNA Technical Leader are as follows:

- Keeps lab Supervisor informed on technical issues
- Oversees QA/QC program for section and keeps proper records (QAS 5.2.3.2.6)
- Maintains proficiency in current methods of analysis
- Oversees proficiency testing of analysts (QAS 5.2.3.2.6)
- Performs technical and administrative reviews (if at one time proficient in the analysis being reviewed)
- Responsible for technical problem solving and analytical methods
- Oversees safety practices
- Makes technical decisions on casework
- If technical issues cannot be resolved, informs supervisor of problem
- Evaluate and document approval of validations of new instrumentation and methods (QAS 5.2.3.2.1).
- Oversees training and evaluates competency of analysts and trainees (including transcripts and training records), and recommends trainees for approval to do casework (QAS 5.2.3.2.2, QAS 5.2.3.2.6)
- Oversees maintenance of equipment and keeps proper records
- Oversees the QA/QC of reagents, kits, and instrumentation
- Recommends continuing education of analysts
- Maintains reference material and library
- Keeps informed on new technologies and legal issues
- Recommends new equipment or facility changes to lab Supervisor

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- Recommends procedure manual updates, keeps SOP current
- Monitors courtroom testimony of examiners
- Review and document the review of internal and external audits and, if applicable, approve corrective action (QAS 5.2.3.2.4)

- Local CODIS Administrator

See appendix 8A in the DNA Technical Manual

- DNA Forensic Scientist

The DNA Forensic Scientist will have a B.A./B.S. degree or its equivalent in a biology, chemistry, or forensic science related area. The DNA Forensic Scientist will also have successfully completed a minimum of 3 college courses in the areas of Biochemistry, Genetics, and Molecular Biology totaling a minimum of 9 credit hours. College course work or training in the subject area of statistics and/or population genetics is also required.

The DNA Forensic Scientist will be responsible for preservation, characterization, collection, documentation, and DNA typing of evidence while following guidelines contained within the DNA Section SOP. The DNA Forensic Scientist will also be available to testify in court to present the facts obtained within a case. The DNA Forensic Scientist is responsible for maintaining the chain of custody of the evidence while it is in their possession. The DNA Forensic Scientist will stay proficient in all disciplines in which casework samples are being processed as required by the FBI DNA Quality Assurance Audit Document. The DNA forensic scientist will help with validation, training, QC, reagent preparation, etc. as required by the DNA Supervisor and/or Technical Leader.

- DNA Forensic Scientist Senior

The DNA Forensic Scientist Senior will meet all the requirements of the DNA Forensic Scientist as well as have three years forensic DNA casework experience, not including training. The three years of experience will include all aspects of the typing process from extraction to interpretation with report signing capabilities. Additional requirements set forth by Human Resources may apply.

- Serologist

The Serologist will have a B.A./B.S. degree or its equivalent in a biology, chemistry, or forensic science related area.

The Serologist will be responsible for preservation, characterization, collection, and documentation of evidence while following guidelines contained within the DNA Section SOP. The Serologist will also write reports and be available to testify in court to present the facts obtained within a case. The Serologist is also responsible for maintaining the chain of custody of the evidence while it is in their possession. The Serologist will stay proficient in methods in which casework samples are being processed.

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- Technician

The technician will have a minimum of 48 college credit hours to include a minimum of 8 biology or chemistry credit hours.

The technician will perform laboratory duties to include reagent preparation, equipment QC, validation, and other lab maintenance duties as needed. The technician may also be responsible for preservation, presumptive testing, collection, and documentation of evidence while following guidelines contained within the DNA section SOP. The technician will also maintain the chain of custody of the evidence while it is in their possession and be available to testify in court. The technician will pass a competency test for the techniques he/she will perform on evidence items. The technician will not interpret data, reach conclusions on typing results, or prepare final reports and will be supervised by a qualified analyst.

- Laboratory Technical Support Personnel

Laboratory technical support personnel will have a minimum of a high school degree.

Laboratory technical support personnel will perform laboratory duties exclusive of analytical techniques on forensic evidence. The duties will include reagent preparation, equipment QC, validation, and other lab maintenance duties as needed. They will have documented training specific to their duties (QAS 5.7).

1.14 Planning and Development

No Supplemental Requirements

1.15 Purchasing Supplies and Services

No Supplemental Requirements

1.16 Management Review System

No Supplemental Requirements

1.17 Equipment and Supply Inventory

No Supplemental Requirements

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2 Facility Design and Security (QAS 3.1.1.4, QAS 6.1)

2.1 DNA Section Physical Plant/Space and Design

The Serology/DNA laboratory will have space for evidence examination, DNA extraction, PCR setup, and amplified DNA product. Refer to Appendix 2A Floor Plan for the location of these areas.

The evidence examination area, DNA extraction area, and PCR setup area will be separate from each other. This can be accomplished by maintaining separate physical spaces for each task or by conducting these tasks at separate times (QAS 6.1.2, ISO 5.3.3). If conducted in the same space at separate times, the space will be decontaminated between tasks.

The amplified DNA product area will be physically separate from all other areas. Entrances to the amplified product area will have a door (QAS 6.1.3).

- Evidence Examination Area

The examination of evidence will primarily be performed in the screening rooms. Equipment in the main lab, such as ovens, centrifuges, and microscopes, may also be used. The tasks performed will include all screening, trace evidence collection (if applicable), body fluid identification testing, selection and cutting of stains, and body fluid extraction for serological tests.

- DNA Extraction Area

The extraction of known and questioned evidentiary samples will be performed in the DNA extraction area. The tasks performed will include DNA extraction, purification, and concentration. Microscopy may also be performed in this area.

- Quantitation Setup Area

Quantitation setup will be performed in the quantitation setup area.

- PCR Setup Area

The setup of PCR amplification reactions will be performed in the PCR setup area. All amplification setup steps including adding template DNA will be performed in the PCR setup area. A laminar flow hood or PCR setup hood dedicated to amplification setup is recommended. A UV light may be run after setup.

- Amplified DNA Product Area

The generation, analysis, and storage of amplified DNA product will be in the amplified DNA product area. Once amplified, no samples will leave the amplified DNA product area unless securely packaged. Equipment, reagents, and supplies in the amplified product area are dedicated and will not be removed unless properly decontaminated.

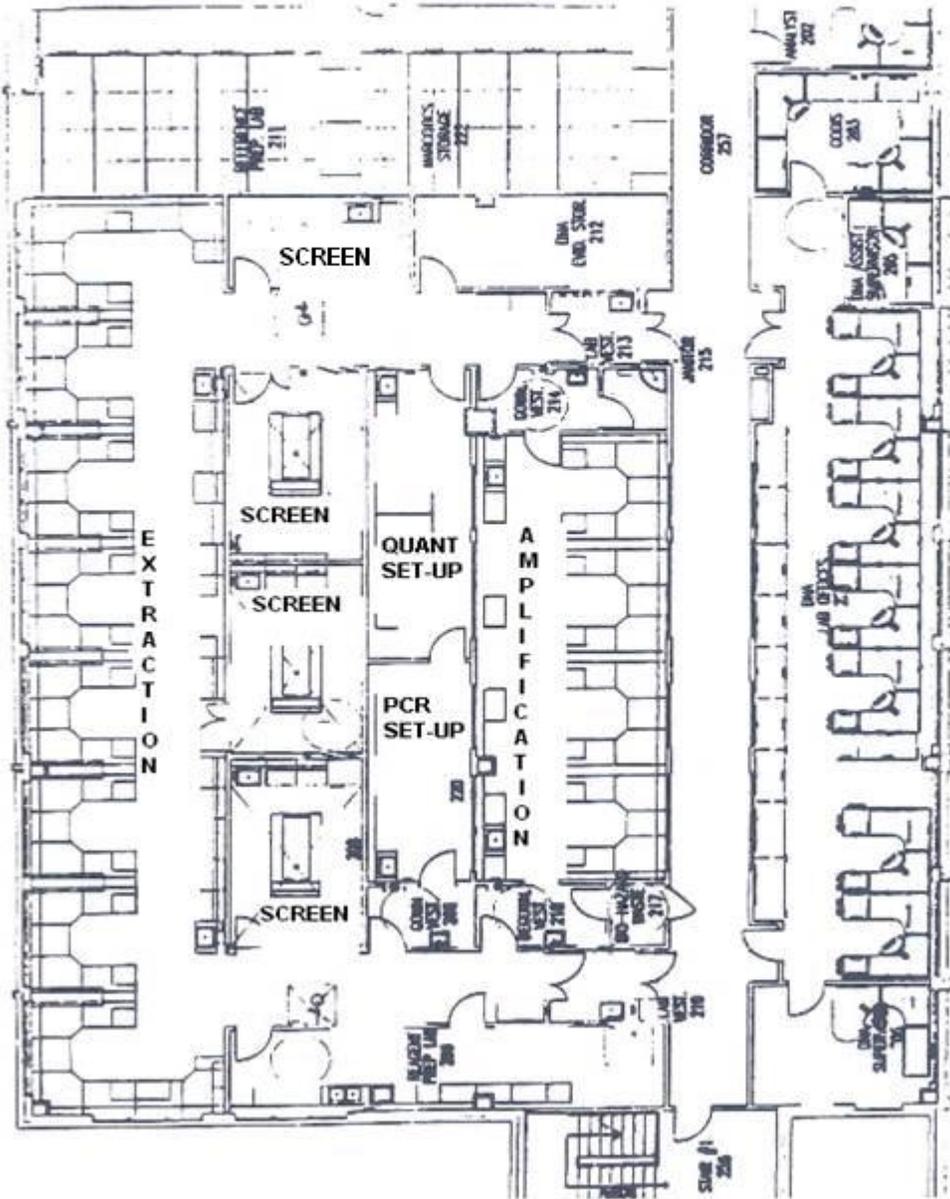
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2.2 Security

Short term storage of evidence and cuttings/swabs in the DNA section will be in the screening rooms. These doors will be locked when the laboratory is unoccupied. Long term storage of evidence will be in the evidence storage room and will include the walk in freezer, supplemental freezer, and the evidence shelves. The door to this room will be locked when the laboratory is unoccupied. DNA extracts will be stored short term in refrigerators in the extraction lab during processing. After processing, they will be stored in the walk in freezer. Evidence swabs and cuttings will be stored short term (during processing) in the screening room or extraction area refrigerators and will then be stored in the walk in freezer for long term storage (QAS 7.1.4).

Appendix 2A

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3 Quality Assurance

DNA Standard Operating Procedures
Effective Date: January 1, 2014

Approved by Laboratory Director
Printed Copies are not Controlled
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3.1 Proficiency Testing (QAS 3.1.1.11)

External DNA proficiency tests will be administered to DNA analysts as follows: there will be two tests per calendar year with one test in the first six months of the calendar year and one test in the last six months of the calendar year. The interval between consecutive tests must be at least four months and not to exceed eight months (QAS 13.1). The submitted date of the proficiency test will serve as the date for calculation of the aforementioned time frame (QAS 13.1.3). All CODIS core loci will be attempted for all samples on DNA proficiency tests (QAS 13.1.5). In addition, the DNA Technical Leader will maintain a copy of the analysis documentation for each proficiency test as well as any documentation of discrepancies/errors and subsequent corrective actions (QAS 13.1.7.3). Newly qualified analysts will be proficiency tested within 6 months of the date of their qualification (QAS 13.1.2). Each analyst will complete their own proficiency test (QAS 13.1.4).

All final reports will be graded as satisfactory or unsatisfactory (QAS 13.1.7.4). A "Satisfactory" rating on the Proficiency Review Form (FSD 009) indicates that all reported inclusions are correct, all reported exclusions are correct, and all reported genotypes and/or phenotypes are correct according to consensus results or within the laboratory's interpretation guidelines (QAS 13.1.7). A "Satisfactory" rating will only be applicable when no analytical errors were observed for the DNA profile typing data (QAS 13.1.7.4.1). The Technical Leader will sign or initial the Proficiency Review Form to indicate knowledge of the results (QAS 13.1.9).

Any results that are reported as inconclusive or not interpretable will be consistent with the laboratory's interpretation guidelines and will be reviewed by the technical leader (QAS 13.1.7.2, QAS 13.1.7.2.1). Any administrative errors and corrective actions pertaining to the report will be documented (QAS 13.1.7.4.1.1)

The following records will be maintained: test-set identifier (QAS 13.1.6.1), identity of the analyst (QAS 13.1.6.2), date of analysis and completion (QAS 13.1.6.3), copies of all data and notes supporting the conclusions (QAS 13.1.6.4), proficiency test results (QAS 13.1.6.5), any discrepancies noted (QAS 13.1.6.6), and any corrective actions (QAS 13.1.6.7).

The Technical Leader will inform the CODIS administrator of all non-administrative discrepancies that affect the typing results and/or conclusions at the time of discovery.

Individuals using both manual and automated methods will be proficiency tested on at least one manual method and one automated method per year (QAS 13.1.1).

3.2 Court Testimony Monitoring

No Supplemental Requirements

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3.3 Case Review (QAS 3.1.1.10)

All case files and reports will be technically reviewed (with the exception of information only reports) and administratively reviewed (**QAS 12.1**). Technical review will consist of the review of the entire case file as well as reanalysis of the electronic data (**QAS 12.2.1**). The technical reviewer will make a table of their analysis results of profiles used for interpretation and will compare those results to the results on the electropherograms in the case file. The technical reviewer will document on their table the run date, the date of review comparison, and that the results agree (**QAS 12.2.2**). The technical reviewer will also identify on the table any inconclusive loci or profiles to show agreement with the call (**QAS 12.2.3**). Inclusions and exclusions will also be verified by the technical reviewer (**QAS 12.2.3**). Administrative review will include the entire case file and report (reanalysis of data not required)(**QAS 12.3**). Chain of Custodies will be reviewed during the review process by the technical and administrative reviewers. The Serology/DNA Review Form (DNA 015) will be used to document technical and administrative review completion. Each criterion to be evaluated during the technical review is listed on the Serology/DNA Review Form (DNA 015)(**QAS 12.2**). "NA" will be used for criteria not relevant to a particular case. Examples are as follows (but not limited to):

- Check stochastic threshold – Swab solution only cases
- Major/Minor component ratio (60%) – cases that do not meet criteria
- CODIS sheet checked - cases with no CODIS entry
- Single source statistics checked – No stats or mixture stats only
- Mixture statistics checked – No stats or single source stats only
- Inclusions/exclusions called properly – cases with no reference samples
- Balanced peaks-Single Source Qs w/ stats – Mixtures or single source with no stats
- Reasons for reinjections noted – cases with no reinjections

Any discrepancies between the analyst and technical reviewer that cannot be resolved will be handled by the Technical Leader (QAS 12.5). During administrative review, the count sheets will be removed from the case folder and stored in the Count Sheet Binders.

Technical reviewers will be employees or contract employees of the DNA laboratory and will be current or previously qualified analysts in the methodologies being reviewed. Each technical reviewer will have successfully completed a competency test in the relevant DNA technology prior to performing technical reviews and will participate in the external proficiency testing program of the DNA laboratory to the extent they participate in the review of DNA data (QAS 5.5, QAS 5.5.1, QAS 5.5.2, QAS 5.5.3, QAS 12.1.1).

- Review guidelines:
 - Variability in the case record documentation is allowed as long as the required SOPs (Division and DNA) and Technical Manual guidelines are followed, and the information is clear, concise, and accurate.
 - If a case is returned to an analyst or reviewer due to corrections, the analyst and/or reviewer will re-initial and re-date the Review Form (DNA 015)
 - Highlighting is optional
 - If the statistics are for the entire profile (all above threshold alleles), the initials of the included individuals are not required on the statistics print out.

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- "An attempt to extract DNA" may be used but is not required when no profiles are obtained.
- Capitalization inconsistencies of words that are not names is acceptable. For example, "Item" and "item".

If a **technical** change is made to **examination** records during the administrative review, the folder will be returned to the technical reviewer who will re-date and re-initial the technical reviewer line on the review form. The folder will then be returned to the administrative reviewer for completion.

3.4 Laboratory Audits (QAS 3.1.1.13)

The DNA section will undergo an annual audit using the FBI DNA Quality Assurance Audit Document. Every other year a qualified auditor (QAS 15.2a) from an external agency must conduct the audit (QAS 15.2). This individual must currently or previously been qualified in the current DNA technology and platform (QAS 15.2b). Audits must be conducted once per calendar year, with the interval between audits not less than six months and not exceeding 18 months.

For internal audits, the audit will be conducted by a team which includes an individual trained by the FBI in auditing using the Quality Assurance Standards (QAS 15.1a, QAS 15.3a) that is currently or previously qualified in the current DNA technology and platform (QAS 15.1b, QAS 15.3b).

Documentation will be maintained showing which individuals have had their education, experience, and training qualifications evaluated and approved (minimum of 2 external audits)(QAS 15.2.1). Documentation will also be maintained of the validations evaluated and approved (minimum of 1 external audit) (QAS 15.2.2).

Audits will be conducted using the current version of the FBI DNA Quality Assurance Standards (QAS 15.4) and the audit documents, including any corrective actions, will be reviewed by the technical leader (QAS 15.5). All external audit documentation and laboratory responses, if applicable, will be forwarded to the FBI within 30 days of the receipt of the documents at the laboratory (QAS 15.5.1). Previous audit documents will be retained and available for review (QAS 15.6).

3.5 Validation (QAS 3.1.1.6, 8.1)

Developmental validation studies will have been performed (by the manufacturer or by another laboratory) prior to use of a technology (QAS 8.2). Citations and publications of such will be maintained (QAS 8.2.1). Peer reviewed publications of a technology will also be available (QAS 8.2.2).

Internal validations on new instrument models or technologies (including change in test kit or platform QAS 8.3.3) to the lab will include the following when applicable: known and non-probative evidence samples or mock samples, reproducibility and precision, sensitivity and stochastic studies, mixture studies, and contamination assessment (QAS 8.3.1).

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Quality assurance parameters and interpretation guidelines, including as applicable, guidelines for mixture interpretation, will be defined pursuant to internal validation (QAS 8.3.2).

Validations and modified procedures must be approved by the Technical Leader (QAS 8.3).

3.6 Instruments and Equipment (QAS 3.1.1.8, QAS 10.1)

Whenever substantial changes are made to a protocol or when material modifications occur, the process will be subjected to an appropriate internal validation including, but not limited to, evaluation using an appropriate NIST or NIST-traceable reference material (minimum of 3 samples) or a minimum of 3 mock samples.

Internal validation of all new manual and robotic methodologies will be conducted and reviewed and approved by the technical leader prior to use on casework. Internal validations will include (when applicable): known and non-probative evidence samples or mock evidence samples, reproducibility and precision, sensitivity and stochastic studies, mixture studies, and contamination assessment. Necessary interpretation guidelines will be developed based on the validation results.

The Technical Leader is responsible for evaluating all methods used by the laboratory and for proposing new or modified analytical procedures to be used by the examiners. Technical Leaders may engage in or direct such activities within their laboratories. No new or modified method, i.e., any method not already described in the SOP, is to be used without the documented approval of the Technical Leader.

The laboratory will annually check its DNA procedures against an appropriate and available NIST standard reference material or standard traceable to a NIST standard (at a minimum, from amplification to characterization)(QAS 9.5.5).

- **Critical Equipment (QAS 10.2, QAS 10.4)**

The following critical equipment must be maintained and subjected to quality control measures (annual performance check and performance check after repair and/or service as well as of new equipment): calibration thermometer (QAS 10.2.1.1), ABI Prism 3130s (QAS 10.2.1.7, QAS 10.4.1.3), pipettes (QAS 10.2.1.8), balances (QAS 10.2.1.2), thermal cyclers (QAS 10.2.1.4, QAS 10.4.1.4), RT PCR quantitation instruments (QAS 10.2.1.4, QAS 10.4.1.4), thermal cycler temperature-verification system (QAS 10.2.1.3), and extraction and liquid handling robots (QAS 10.2.1.6, QAS 10.4.1.2). The annual performance check documentation will be kept in the performance check notebook, equipment notebook, or equipment calibration notebook. Any after service or repair documentation will be kept in the equipment's notebook or equipment calibration notebook.

- **Non Critical Equipment**

Additionally, the following non-critical equipment will be maintained as outlined below: water baths, microcentrifuges, refrigerators/freezers, equipment thermometers, and hoods.

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Procedures (QAS 10.3)

Guidelines for general equipment are contained within the Forensic Science Division SOP. Below are the procedures for equipment specific to the DNA section.

- **Water Bath**

Water baths are dedicated equipment whose temperature is routinely maintained at ~56°C for DNA procedures.

- Observed Temperature

Observe the temperature reading on the thermometer or via the electronic temperature monitoring program (should be recorded weekly). The temperature should be within +/- 1°C of expected temperature. If not within the acceptable range, use the control knobs to adjust the temperature to the acceptable range. It may be necessary to adjust the digital display. If the equipment is monitored electronically and is out of range, a reading can be taken with a currently calibrated thermometer to verify temperature. If the temperature is not stable, have the water bath repaired or replace the water bath.

- Water Condition

The water in the bath should be clear and clean with no evidence of bacterial/fungal growth or rust. If the water becomes dirty, discard and clean the water bath. Replenish with water and document on the log.

- Form: 032

- **Pipettes**

Please see the FSD SOP for general requirements. DNA forms 047 and 048 will be used.

- **Microcentrifuges**

Microcentrifuges are bench top, unrefrigerated centrifuges that have been designed for centrifugation of tubes. These microcentrifuges are equipped with fixed angle rotors. The maximum speed is specified in the operations manual for each centrifuge.

- Quality Control Procedure

- RPM

The speed as measured by a tachometer must correspond to a predictable number on the speed control setting or the digital readout. If the two speeds do not correspond within 10%, the centrifuge must either be replaced or repaired. This quality control procedure is performed once every six months.

- Maintenance Procedure

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Centrifuge housing, rotor chamber, and rotor accessories should be cleaned with neutral cleaning agents as needed; typically as part of the lab's cleaning schedule. All parts must be dry prior to use.

➤ Form: 012

- **Refrigerators/Freezers**

Each refrigerator/freezer used in the laboratory has been shown to be capable of maintaining the optimum temperature range required for storing reagents and samples. A thermometer for monitoring the temperature has been placed in the upper/lower compartments unless a digital thermometer is built into the unit or is monitored electronically. Refrigerators/freezers are monitored during working hours to ascertain that they are functioning. Weekly records of temperature readings should be maintained. No food or beverages are allowed in the laboratory refrigerators.

➤ Quality Control Procedure

- Temperature

The refrigerator maintains optimum temperature with the required range of 0°C - 8°C. The freezer compartment should maintain a temperature below 0°C. Independent freezers should maintain a temperature below -10°C. The temperatures also need to be appropriate for the reagents stored within. If the temperature is not being maintained at the prescribed values, verify that the temperature regulation control is at the proper setting and adjust if necessary. Any adjustments should be documented on the log. It may also be necessary to reduce over-crowding in an effort to increase air circulation in all compartments. If necessary, a technical representative may need to be called for service or the refrigerator/freezer may need to be replaced if repair is not adequate. If the equipment is monitored electronically and is out of range, a reading can be taken with a currently calibrated thermometer to verify temperature.

➤ Maintenance Procedure

Each refrigerator/freezer is maintained in working order and repaired or replaced if necessary. The compartments are kept clean and well organized.

- **Electronic Temperature Monitoring System**

Refrigerators, freezers, water baths, ovens, heat blocks or other equipment may be monitored via the Andover Controls' electronic monitoring system which uses Contium building automation software. This program is maintained by APD Facilities Maintenance.

➤ Quality Control Procedure

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Each probe will be compared against a currently calibrated digital thermometer at least once every two years and must be +/- 1.0°C.

➤ Maintenance Procedure

If the electronic temperature falls outside the acceptable range, adjustments will be made by building maintenance personnel or the contracted company or the probe can be replaced by building maintenance personnel.

• **ABI Prism 3130 Genetic Analyzer**

The ABI Prism 3130 Genetic Analyzer is a capillary electrophoresis instrument used to separate DNA fragments based upon size and fluorescent tags.

➤ Quality Control Procedure

A performance check will be conducted annually and after service or repair to ensure the ABI Prism 3130 Genetic Analyzer is working properly and will consist of an allelic ladder, positive control, and negative control analyzed under normal conditions to ensure all peaks are being called appropriately and that the negative control contains no above threshold peaks. The GeneMapper ID™ data from this run must be printed and placed in the appropriate logbook. If the instrument fails the performance check, it will be taken out of service until it is repaired and passes the performance check.

A new spectral should be made once every 6 months or as needed for each instrument in the laboratory. Follow the manufacturer's guidelines for making the spectral and verifying its accuracy.

➤ Maintenance Procedure

The lab has a Planned Maintenance agreement with Life Technologies for the maintenance of the Genetic Analyzer. This plan allows for 1 planned-maintenance visit per year by a Field Service Engineer. After the planned maintenance, a ladder, positive control, and negative control must be run and analyzed as described above. The GeneMapper ID™ data from this run must be printed and placed in the appropriate logbook.

The instrument components should be cleaned as needed. The reagents should be replaced or replenished as necessary. The lot numbers of the reagents will be documented on the reagent log and cleaning will be documented on the cleaning log.

Analytical data should be routinely archived and the hard drive should be analyzed with the system disc defragmenter as necessary.

• **Thermal Cyclers**

Thermal Cyclers automate the polymerase chain reaction (PCR) for amplifying DNA.

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➤ Quality Control Procedure

A performance check including temperature calibration, temperature uniformity, and diagnostic tests must be performed once every 6 months and after repair or service. Follow manufacturer's instructions for performing these tests. Any tests that result in a "failed" notification by the instrument will necessitate recalibration or repair of the instrument by the manufacturer or a qualified service technician. Calibration log sheets will be maintained and available for inspection.

➤ Maintenance Procedure

The Thermal Cycler is an instrument that is located in a dedicated portion of the laboratory. The sample block and exterior surfaces should be cleaned approximately once every 6 months.

➤ Form: 019

• **Temperature Verification System**

The temperature verification system is used to perform the thermal cycler temperature calibrations. It will undergo an annual calibration by an outside vendor. Documentation will be maintained in the thermal cycler log book.

• **Hoods**

The hood, when used with proper technique, is effective in reducing the potential for exposure of both product and personnel to airborne biological or particulate chemical agents

➤ Quality Control Procedure

The hoods should be re-certified at least once a year.

➤ Maintenance Procedure

The hoods are maintained by the Building Services unit.

• **Water Filtration System**

The water filtration system is an integral part of the lab facility. It will be monitored and maintained by the Building Services unit. Stand-alone units in the laboratory have a digital display and can be used when the reading is ≥ 18 MOhm·cm. If the reading is below 18 MOhm·cm, the unit will not be used and a service technician will be contacted.

• **ABI 7500**

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The ABI Prism 7500 Sequence Detection System is a real-time PCR process used for quantitation of DNA. The system consists of the 7500 instrument and a computer with appropriate software.

➤ Quality Control Procedure

The standard curve for each run will be evaluated. The approximate requirements include a slope of -2.9 to -3.35 and a R^2 value ≥ 0.98 . Variations can be signed by the Technical Leader or technical reviewer.

➤ Maintenance Procedure

A performance check will be conducted annually and after service or repair to include the system test (all tests must "pass"), contamination check (all wells must have readings below 1200), and the temperature verification check. The lab has a Planned Maintenance Agreement with Applied Biosystems for the maintenance of the ABI 7500. This plan allows for 1 planned-maintenance visit per year by an Applied Biosystems Field Service Engineer. The service engineer will perform the temperature verification check and ensure it meets the specifications. If any wells are identified as having a reading over 1200 in the contamination check, the wells will be cleaned with diH_2O or ethanol and the test (and cleaning, if needed) will be repeated until the values are below 1200.

Any repair or service must be documented and maintained in the appropriate logbook.

The following periodic checks may also be run and documented:

- Background Assay
- Lamp check
- ROI
- Spectral Calibration

➤ Form: 008

• **Extraction Robots**

The QIAcube extraction robots and Maxwell 16 extraction robots will be maintained according to manufacturer guidelines.

➤ Quality Control Procedure

Ensure appropriate protocols are used and that each run completes as expected.

➤ Maintenance Procedure

- Robots will be cleaned as necessary and per manufacturer instructions.

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- After repair or service, a performance check that includes a water run or mock sample run will be performed to ensure the robot is working appropriately by the completion of the run. If the run does not complete appropriately, the instrument will be taken out of service until it passes the performance check.
- An annual performance check will also be documented as described above.

• **CAS-1200 Liquid Handling Robot / QIAgility Liquid Handling Robot**

The Corbett CAS-1200 Liquid Handling robot and the QIAgility are compact and precise liquid handling systems that will be used to set up quantitation standards and samples, amplification reactions, and CE runs.

➤ **Quality Control Procedure**

Ensure appropriate protocols are used and that each run completes as expected or is documented as to what issues arose and how they were resolved.

➤ **Maintenance Procedure**

- Robots will be cleaned as necessary and per manufacturer instructions.
- An annual PM should be performed by QIAgen.
- The integral UV light should be turned on after each run. The bulb is replaced annually by QIAgen during the robot's PM.
- After repair, a performance check that includes a mock run will be performed to ensure the robot is working appropriately. An acceptable standard curve, per policy, shows proper operation (Corbett CAS-1200) or correct results from a positive control and negative control (QIAgility).
- An annual performance check will also be documented using the criteria above.
- If the instrument does not pass the performance check, it will be taken out of service until it passes.

3.7 Reagents (QAS 9.2)

Preparation of in-house reagents will be documented on the Reagent Preparation Form (DNA 010). All component's names, lot #s, and expiration dates will be tracked on the form.

Critical Reagents (QAS 9.3)

The following are critical reagents for the DNA Laboratory and require a QC check prior to use on casework samples:

Life Technologies: Quantifiler

Promega: DNA IQ Casework Sample Kit, Swab Solution kit, PowerPlex Fusion kit

QIAgen: QIAamp® DNA Investigator Kit

- **Amplification Kits (Fusion) (QAS 9.3.1b).**

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Each new lot of an amplification kit must be subjected to an internal quality control test as outlined below:

- The positive control DNA must be run to determine its activity. A full correct profile must be achieved.
- An amplification blank must be run to determine its purity (i.e., no contamination in reagents). No detectable alleles may be present above threshold.
- All reagents in the kit must be used to demonstrate their viability.
- The allelic ladder must be run to determine that all of the appropriate alleles are detected.

If any component of the kit does not meet the aforementioned criteria the process will be repeated. If the kit fails the QC a second time the Technical Leader will be informed and the kit will not be used on casework samples.

- Form: 009

- **Quantifiler Kits (QAS 9.3.1a)**

Each new lot of Quantifiler kits must be subjected to an internal quality control test as outlined below:

- A set of standards will be prepared and run according to procedure along with a template control.
- The R^2 value will be evaluated and must be ≥ 0.98 .
- The slope value must be within the range of -2.9 to -3.35.

If the kit does not meet the aforementioned criteria, the process will be repeated. If the kit fails the QC a second time the Technical Leader will be informed and the kit will not be used on casework samples.

- Form: 023

- **QIAamp[®] DNA Investigator Kit**

Each new lot of QIAamp[®] DNA Investigator kits must be subjected to an internal quality control test as outlined below:

A known blood or saliva sample and a reagent blank will be processed through the extraction kit to check the quality of the reagents.

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The DNA extracts will be quantitated (RB optional), amplified, and analyzed to ensure the correct profile was produced and there are no detectable alleles in the RB above threshold (75 RFU for RB).

If the kit does not produce the aforementioned results, the samples will be re-extracted and re-analyzed. If the kit fails the QC a second time the Technical Leader will be informed and the kit will not be used on casework samples.

➤ Form: 042

- DNA IQ Casework Sample Kit

Each new lot of DNA IQ kits must be subjected to an internal quality control test as outlined below:

A known blood or saliva sample and a reagent blank will be processed through the extraction kit to check the quality of the reagents.

The DNA extracts will be quantitated (RB optional), amplified, and analyzed to ensure the correct profile was produced and there are no detectable alleles in the RB above threshold (75 RFU for RB).

If the kit does not produce the aforementioned results, the samples will be re-extracted and re-analyzed. If the kit fails the QC a second time the Technical Leader will be informed and the kit will not be used on casework samples.

➤ Form: 040

- Promega Swab Solution Kit

Each new lot of Promega Swab Solution kits must be subjected to an internal quality control test as outlined below:

A known blood or saliva sample and a reagent blank will be processed through the extraction kit to check the quality of the reagents.

The DNA extracts will be amplified, and analyzed to ensure the correct profile was produced and there are no detectable alleles in the RB above threshold (75 RFU for RB).

If the kit does not produce the aforementioned results, the samples will be re-extracted and re-analyzed. If the kit fails the QC a second time the Technical Leader will be informed and the kit will not be used on casework samples.

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➤ Form: 041

3.8 Document Management

The DNA Technical Leader will maintain the following records: Proficiency test results (supporting documentation, corrective action reports, and proficiency review forms); casework corrective actions, internal and external audits using the DNA audit document; and training records and competency tests. The Quality Assurance Manager will maintain ASCLD/LAB audit documentation. Continuing education certificates and/or agendas and court testimony monitoring forms will be maintained in the Analyst Notebooks. Case files will be stored in the file room (QAS 3.2).

Analyst notebooks will contain the following: Transcripts, CV, Continuing Education Certificates, Approval for Casework Forms, Proficiency Testing Review Forms, Court Testimony Monitoring Forms, and professional affiliation documentation (QAS 5.1.4).

The quality system review as applicable to DNA will be reviewed annually and will be performed under the direction and documented approval of the Technical Leader (QAS 3.3, QAS 5.2.3.2.5, QAS 9.1a). This review includes laboratory procedures in the SOP, technical manual, and training manual.

3.9 Deviation from Documented Procedures

No Supplemental Requirements

3.10 Preventive and Corrective actions

Prior to implementation, all corrective actions will have the documented approval of the Technical Leader **(QAS 14.2)**.

3.11 Suggestions/Complaints

No Supplemental Requirements

3.12 Customer Survey

No Supplemental Requirements

3.13 Reference Standards/Materials

No Supplemental Requirements

3.14 Reference Collections and Databases

No Supplemental Requirements

3.15 Examination Verification

DNA Standard Operating Procedures
Effective Date: January 1, 2014

Approved by Laboratory Director
Printed Copies are not Controlled
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No Supplemental Requirements

3.16 Contamination Detection and Prevention (QAS 9.7)

Contamination is defined as inadvertent transfer of DNA from one sample to another, from a person to a sample, or from a person or sample to bulk reagents or consumables. Any and all above threshold contamination events (minimum threshold used within a case) that occur within the DNA section will be summarized in an incident log that will document the details of the contamination incident including steps taken toward identifying the source or step in the process where the contamination occurred and the cases affected by the contamination. This documentation will be used in lieu of a QIN. Documentation will also be maintained in the files of affected cases. An incident log tracking form will also be completed. In lab (DNA lab) contamination will typically not be used for interpretation of the affected samples. If the samples cannot be reprocessed from the step where the contamination is deemed to have occurred, the samples will typically be called inconclusive. In the event the reagent blank shows contamination but samples within a case that are deemed uninterpretable do not show signs of contamination, the analyst may report the uninterpretable results without reprocessing.

Exceptions may be made by the technical leader allowing for use of samples with indications of contamination. Documentation by the technical leader is required in these instances.

Contamination detected in the DNA section which occurred from a prior lab source (i.e., other section personnel) will be handled per current division guidelines. Any non-lab personnel detected or included in a sample will be listed as elimination samples and the sample can be interpreted.

Samples can become contaminated with DNA from the environment, from other samples during sample preparation, or from amplified DNA product from a previous amplification. Reagent blanks and negative amplification blanks are used to detect contamination.

Contamination will be suspected and investigated whenever more than two alleles appear at a locus when the sample is believed to be of one source (unless consistent with a tri-allele) or whenever a negative control or reagent blank yields peaks above the minimum analysis threshold for the batch. In addition, contamination may be suspected and investigated under other circumstances at the discretion of the examiner, Technical Leader, or Supervisor.

- Prevention and decontamination

10% bleach, a commercial DNA decontaminant or ethanol will be used as a decontaminant. Ethanol is effective in decontaminating surfaces of microbial agents and wiping away particles but will not adequately destroy DNA on surfaces or equipment.

Wear disposable gloves and lab coats during all testing. Masks are also highly recommended. Change gloves frequently and whenever gloves may have become contaminated. Discard gloves when leaving a work area. Centrifuge all liquid to the bottom of closed microcentrifuge tubes before opening. A de-capper may be used. Use sterile, disposable pipette tips and microcentrifuge tubes. Use aerosol-resistant pipette

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tips while working with any sample that may be amplified. Change pipette tips between samples.

In the evidence examination area, clean work surfaces thoroughly with decontaminant at least at the end of each evidence examination session. Use disposable bench paper whenever possible and change at least at the end of each evidence examination session. Use a clean cutting surface such as weighing paper for each piece of evidence. Protect supplies of this paper from dust and other particulates or aerosols. Clean instruments (scissors, forceps) between evidence samples. Alternatively, use a fresh scalpel blade with each sample. To prevent contamination of other standards or evidence, handle liquid samples such as a blood standard one at a time and with no other evidence open in the vicinity.

In the DNA extraction area, clean work surfaces thoroughly with decontaminant at least at the end or beginning of each DNA extraction session. Limit talking during sample handling and wearing a mask is highly recommended.

In the PCR setup area, add DNA template last to the PCR setup tubes to minimize inadvertent transfer between setup tubes and resultant cross contamination. Limit talking during sample handling. It is recommended that the lab irradiate work surfaces and equipment in the PCR setup area with ultraviolet germicidal lamps. Surfaces not irradiated will be treated with decontaminant.

In the amplified DNA product area, wear a dedicated, disposable lab coat when handling amplified samples. Do not wear the lab coat outside the amplified DNA product area. These lab coats will be disposed of when necessary.

Each analyst's DNA profile will be developed for all systems currently in use.

Visitors to the DNA lab will also wear masks, gloves, and lab coats except in the amplification room. If a visitor needs to visit the amplification room and other parts of the lab, they will visit the amplification room last.

- Response to contamination

Any suspected contamination incident must be immediately brought to the attention of the Technical Leader. The Technical Leader may also be required to inform the laboratory supervisor. The Technical Leader will define and direct investigative actions. All actions will be documented, and summary documentation will be retained in the incident logbook.

An example action plan proposal may include the following:

- Repeat portions of the procedure (extraction to typing, if necessary) for the set of samples in which contamination was detected.

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- If no contamination is present, no further action is required. If contamination is still evident in the controls or samples, DNA casework will be discontinued immediately until the source of the contamination is uncovered.
 - Suspected buffers and prepared reagents will be discarded; reagent bottles thoroughly cleaned, decontaminated and autoclaved; and fresh reagents and buffers prepared.
 - The work areas, glassware, pipettes, etc., will be thoroughly cleaned and decontaminated.
 - If necessary, a known sample set will be re-extracted, re-amplified, or re-typed (depending on the nature of the contamination) using fresh reagents. If no contamination is present, casework will be resumed.

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APPENDIX 3.15A DECONTAMINATING PROCEDURES (QAS 6.1.5)

Equipment

PCR Setup Hoods	Integrated UV light
Cas-1200	Integrated UV light
QIAcubes	Per manufacturer instructions; approximately monthly or as needed
Maxwell 16s	Per manufacturer instructions; approximately monthly or as needed
9700 Thermal Cyclers	Wipe blocks and exteriors approximately every 6 months
7500	Block contamination check approximately once a month
3130	As needed per manufacturer instructions
Water baths	Replace or add dH ₂ O as needed
Centrifuges	Done as part of general lab cleaning
Pipettes	Wipe shaft as needed with dH ₂ O or DNA Away

General Laboratory Cleaning

A cleaning log has been placed in the laboratory. Regular cleaning is to be done on the bench tops and floors and is to be documented on the log. DNA Away is also available for cleaning work surfaces.

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Appendix 3A

Approved Vendors and Suppliers

- **Approved Vendors-Critical Supplies (These vendors have been evaluated via internal validation of the reagents)**
 - Life Technologies (Formerly Applied Biosystems)
 - Quantifiler
 - Promega
 - DNA IQ kit for Maxwell 16
 - Swab Solution Kit
 - PowerPlex Fusion
 - QIAgen
 - DNA Investigator Kit
- **Approved Calibration Service Companies**
 - Essco Calibration Laboratory
 - Control Company
 - Artel
 - Rainin

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4 Laboratory Records

4.1 Case Record (QAS 11.1b, QAS 11.1c)

All analytical documentation will be retained. All hard copy information will be maintained in the case file with the exception of the count sheets which will be stored in binders. All electronic analysis data will be retained on the APD group drive (which is backed up daily) and will periodically be copied to CDs (QAS 11.1b). The data will be electronically stored in folders designated by the year and month of testing. Administrative documentation not in the case file will be stored in the case record in LIMS.

The start date of an examination will be documented on the Review Form (DNA 015).

4.2 Laboratory Reports (QAS 3.1.1.9, QAS 11.1)

The following items specific to the DNA section must be included in every report (**QAS 11.2**):

- Description of DNA methodology (**QAS 11.2.3**)
- Loci analyzed if DNA typing was performed (**QAS 11.2.4**)
- Disposition of evidence (**QAS 11.2.8**)

The DNA section will typically call the first report within a case “Initial” and any reports thereafter “Supplemental”.

Examples of typical casework results follow. Under each bold heading of test results are ways to state the conclusion. Example wording that should be changed according to the case is denoted by *italics*. Not every situation can be represented here, and wording variations that accurately reflect the findings will sometimes be necessary. When in doubt, consult the Technical Leader or Supervisor.

- **Blood examinations**

No visible blood, no presumptive tests performed.
No stains having the appearance of blood were detected on *Item 1*.

Negative presumptive test(s).
A presumptive test for the presence of blood was negative on *item 1*.

Positive presumptive test(s).
A presumptive test for the presence of blood was positive on *item 1*.

- **Semen examinations**

Negative alternate light source of clothing or bedding.
No stains having the appearance of semen were detected on *the victim's shirt*.

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Negative AP screening of clothing or bedding.

A presumptive test for the presence of semen was negative on *the victim's shirt*.

Positive AP, no further testing.

A presumptive test for the presence of semen was positive on the *victim's shirt*.

Negative or weak AP, negative sperm, negative p30 on swab.

No semen was detected on *the vaginal swab*.

Positive AP, negative sperm, negative p30

No semen was detected *on the stain on the crotch of the panties*.

Negative or positive AP, positive sperm, negative p30.

Semen was detected on *the vaginal swab*.

Negative or positive AP, negative sperm, positive p30.

Semen was detected on *the vaginal swab*; however, no spermatozoa were detected.

Negative or positive AP, few sperm, positive or negative p30.

Semen was detected on *the vaginal specimens*, however minimal spermatozoa were detected.

- Negative or positive P30
p30 (a constituent of semen) was (not) detected on *the vaginal swabs*.

- **DNA analysis**

- Results

The desired end of DNA typing is to determine whether a particular person is or is not a potential source of an item of biological evidence.

The results of each comparison will be one of the following:

- No DNA profiles were obtained.
- No conclusive or interpretable DNA profiles were obtained.
 - A reason must be given when using this statement. Examples are:
 - a) ...due to low signal which makes this result inadequate for ANY comparisons to potential reference sample(s) using currently available techniques.
 - b) ...due to excessive below threshold information
 - c) ...due to the presence of information consistent with a non-DNA lab employee
 - d) ...due to a contamination incident

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- The DNA profile of the evidentiary sample cannot be compared because no known sample profile is available.
- The DNA profile of the evidentiary sample is not consistent with the profile of the known sample.
- The DNA profile of the evidentiary sample is consistent with the profile of the known sample.
- The DNA profile of the evidentiary sample is consistent with a mixture of DNA profiles from more than one individual, one or more of whom may be known.

These results must be included in the report, either implicitly or explicitly, for each evidentiary profile generated. It is not necessary for the report to include the DNA profile(s), although the analyst may include this information in the form of a table. If a locus is inconclusive or not responsive to testing, the profile at that locus will not be reported and will not be included in calculations of statistical significance estimations.

➤ Reporting statements (QAS 9.6.4c)

Different report conclusions are used in the case of single source, major component of a mixed source, and mixed source evidentiary profiles. Criteria for categorizing evidentiary profiles are outlined in the Statistics chapter of this manual. Each prescribed statement is detailed below. When a comparison is made for a subset of the loci attempted, e.g., a partial profile, the analyst will add the wording within the brackets “[]” to reflect the loci used for determining the match but not the brackets themselves or the ellipses. Case-specific words should be substituted for the italic typeface in the prescribed statement.

Reporting statements may include a statement of exclusion or inclusion. A significance statement may accompany a statement of inclusion. For significance estimates that are 1 in at least 312 billion for every population group calculated, and matching results are obtained at all loci (excluding DYS391), the significance statement will identify the source. If the source is not identified, the report will state the approximate population of the world (7,000,000,000) at least once; preferably in the first comparison statement where a source is not identified. Analysts have the choice of expressing large numbers using numerals or words.

Statistical significance statements are not required for non-probative stains. Depending on the case scenario, one example might be suspect's blood on suspect's clothing or the victim's type in the epithelial cell fraction on the victim's body swabs. For probative stains, the analyst will include a significance statement for each inclusion.

- Examples of Reporting Statements

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○ Exclusion

An exclusion should be reported using this wording: “The DNA profile from *Item 1, stain 3*, is not consistent with the DNA profile of *Person A*. *Person A* is excluded as the contributor of this profile.”

OR

“The DNA profile from *Item 1, stain 3*, is consistent with a mixture. *Person A* is excluded as a contributor to this profile.”

○ Inclusion - Single source

For evidentiary profiles that meet the single source criteria, an inclusion should be reported using this wording:

The partial DNA profile from *stain* is consistent with the DNA profile of *Person A*. *Person A* cannot be excluded as the contributor of this profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045*. At these loci,] The probability of selecting an unrelated person at random who could be the contributor of this DNA profile is approximately 1 in ___ for Caucasians, 1 in ___ for African Americans, and 1 in ___ for Hispanics.

AND

“Based on these probabilities, *Person A* is the source of this profile (excluding identical siblings).”

OR

“The approximate world population is 7,000,000,000.”

○ Inclusion - Major component of a mixed source

For evidentiary profiles that meet the major component criteria, an inclusion should be reported using this wording:

“The DNA profile from *stain* is consistent with a mixture.”

OR

“The DNA profile from *stain* is consistent with a mixture of *Person A* and *Person B* [and *Person C* and some unknown individual...].”

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AND

“*Person A* cannot be excluded as the contributor of the major component in the profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045*. At these loci,...] The probability of selecting an unrelated person at random who could be the source of the major component in this DNA profile is approximately 1 in ____ for Caucasians, 1 in ____ for African Americans, and 1 in ____ for Hispanics.

AND

“Based on these probabilities, *Person A* is the source of the major component of this profile (excluding identical siblings).”

OR

“The approximate world population is 7,000,000,000.”

- o Inclusion - Mixed source

For evidentiary profiles that do not meet either the single source or major component criteria, a match should be reported using this wording:

“The DNA profile from *Item 1, stain 3* is consistent with a mixture.”

OR

“The [above threshold] DNA profile from *stain* is consistent with a mixture from *Person A* and *Person B* [and *Person C*]. [Additional alleles may be [are] present below threshold]”

OR

The DNA profile from *stain* is consistent with a mixture of at least *number* individuals.

AND

“*Person A* cannot be excluded as a contributor to this profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045*. At these loci,...] The probability of selecting an

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unrelated person at random who could be a contributor to this profile is approximately 1 in ____ for Caucasians, 1 in ____ for African Americans, and

1 in ____ for Hispanics. The approximate world population is 7,000,000,000.”

The determination to use the statement “The DNA profile from *stain* is consistent with a mixture from *Person A* and *Person B* [and *Person C*, etc.]” is based on above threshold alleles. The additional statement "additional alleles are present below threshold" can be used at analyst discretion.

If a match to a major component in a mixture is reported, it is not necessary to also report the statistical significance for the mixture profile. Consider a vaginal swab analysis for example: if the major component of the sperm fraction matches the suspect, it is not necessary to also report the statistical significance of the match of the minor component to the victim.

The term “major contributor” may be used in mixture profiles where there is a more significant donor of DNA that does not meet major component criteria (mixture statistical calculation would be used). This can be made in conjunction with comparisons and does not need to be pre-determined.

Additional phrases such as "the minor component is below threshold" or "additional alleles may be present below threshold" may also be applicable.

The statement about minimum number of contributors should be included in the report. Please see page 56 of the DNA Technical manual for more information.

o Uninterpretable

In some instances, DNA testing may yield uninterpretable results. The following statements may be used. A reason must also be given for uninterpretable profiles:

a) No DNA profiles were obtained from *the shirt stain*.

b) No interpretable DNA profiles were obtained from *the shirt stain* due to *multiple alleles below threshold*.

c) The DNA result from *the shirt stain* was uninterpretable *due to a minimal amount of DNA*.

o Combining statements

For one item of evidence, it may be clearer to combine inclusion, exclusion, and significance statements. This improves readability without changing the meaning.

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Example: The DNA profile from the *stain* is consistent with a mixture. The victim is excluded as a contributor of this profile. William Johnson and John

Williamson cannot be excluded as contributors to this profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045.* At these loci,] The probability of selecting an unrelated person at random who could be a contributor to this profile is approximately 1 in ___ for Caucasians, 1 in ___ for African Americans, and 1 in ___ for Hispanics. Fred Ferguson cannot be excluded as a contributor to this profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045.* At these loci,] The probability of selecting an unrelated person at random who could be a contributor to this profile is approximately 1 in ___ for Caucasians, 1 in ___ for African Americans, and 1 in ___ for Hispanics. The approximate world population is 7,000,000,000.

Similarly, for one item of evidence, it may be clearer to combine inclusion, exclusion, and significance statements for a mixture and a major component of a mixture.

Example: The DNA profile from the *stain* is consistent with a mixture from the victim and the suspect. The victim cannot be excluded as the contributor of the major component in the profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045.* At these loci,] the probability of selecting the DNA profile of the major component is 1 in ___ for Caucasians, 1 in ___ for African Americans, and 1 in ___ for Hispanics. The suspect cannot be excluded as a contributor to this profile. [Statistics were calculated at the following loci: *D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D123391, D19S433, FGA, and D22S1045.* At these loci,] The probability of selecting an unrelated person at random who could be a contributor to this profile is approximately 1 in ___ for Caucasians, 1 in ___ for African Americans, and 1 in ___ for Hispanics. The approximate world population is 7,000,000,000.

- o Alternate reference samples

When no standard reference samples are available, alternate reference samples may be used, e.g., Pap smear, hair from a hair brush, or blood from

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a piece of clothing. When compared to an evidentiary profile, the appropriate prescribed reporting statement will be modified to read:

“The DNA profile from Item 1, stain 3, is consistent with the DNA profile of the Pap smear. Assuming Person A is the source of the Pap smear, Person A cannot be excluded....”

OR

“The DNA profile from Item 1, stain 3, is not consistent with the DNA profile of the Pap smear. Assuming Person A is the source of the Pap smear, Person A is excluded....”

➤ Distribution of Reports

Reports may be emailed or faxed to CODIS laboratories for the purpose of hit disposition as required. Reports that are emailed will be converted to a PDF prior to sending. The analyst faxing a report will confirm the correct number has been entered prior to sending. Any other emailing or faxing will have supervisor approval prior to sending.

4.3 Release of records Information

4.3.1 Release of Information

• Status Updates

It is sometimes necessary to provide status updates to detectives as a case is being processed. When necessary, the following status updates may be provided:

- Presence or absence of apparent blood and/ or semen
- Mere absence or presence of a CODIS eligible profile
- Notification of a CODIS hit that is being verified
- Processing time frames or item presence, appearance, or general characteristics
- Lack of a CODIS match from a keyboard search

4.3.2 Release of Information to the News Media

No additional information

4.3.3 Open Records Request

No additional information

4.3.4 Discovery Orders

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No additional information

4.4 Removal of Records for Court

No Supplemental Requirements

4.5 Archiving Laboratory Case Files

No Supplemental Requirements

4.6 Expunctions

No Supplemental Requirements

4.7 Control of Laboratory Records

No Supplemental Requirements

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APPENDIX 4A LISTING OF OFFENSE CODES

- 01 Homicide
- 01A Attempted Homicide
- 02 Sexual Assault
- 03 Robbery
- 04 Assault
- 05 Burglary
- 06 Theft
- 06C Credit Card Offenses
- 07 Auto Theft
- 08 Arson
- 10 Forgery and Counterfeiting
- 11 Fraud
- 12 Embezzlement
- 13 Stolen Property Offense
- 14 Vandalism
- 15 Weapons Offense
- 17 Sex Offense Other Than Rape
- 18 Controlled Substances Offense
- 18M Controlled Substances Manufacturing
- 20 Offenses Against Family and Children
- 21 Intoxication Offense
- 21D Intoxication Offense (Deceased)
- 22 Liquor Violation
- 23 Drunkenness
- 25 Vagrancy
- 26 Criminal Offense
- 30 Hit and Run
- 31 Traffic Incident
- 32A Questioned Death
- 32B Suicide or Accidental Death
- 33 Non-criminal
- 34 Violation of Probation
- 35 Threatening Correspondence
- 36 Official Misconduct
- 37 Harassment
- 38 Kidnapping
- 39 Officer Involved Shooting

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5 EVIDENCE PROCEDURES

5.1 General Practices

- **Case acceptance and Evaluation**

Before a case is accepted or worked, the case will be evaluated. The examiner should be thoroughly aware of the requested examinations, the reason(s) for the requested analyses, the relevance of the examination in solving the crime or answering certain key questions, and the quality and quantity of the evidence. The work that is performed in the laboratory is usually used to indicate a link between a victim or crime scene and the perpetrator. The results of the examinations may either implicate or exonerate a suspect from involvement in a crime and are meaningful only if the conclusions that are drawn are relevant to the case. Because each case is different, only guidelines can be prescribed; the case evaluation should include consultation with the investigator/prosecutor as necessary.

Once a request for analysis has been received, the Supervisor will typically assign the case to an analyst. If a folder has not been previously created, the analyst will create one.

- **Evidence evaluation**

Before the case is worked, an evaluation should be made to determine the quality and quantity of the evidence that is going to be analyzed. In order to expedite casework, it is recommended that for cases containing large volumes of evidence (excluding sexual assault kits) 5-10 probative items of evidence should be screened. Of those 5-10 items screened, it is recommended that a maximum of 5 evidence stains should continue on to DNA analysis. Additional items/stains may be analyzed at a later date depending on case development and initial DNA analysis results. Decisions have to be made concerning the analytical approach that must be taken to obtain the most useful information. It is often helpful to consult with another qualified examiner, the Technical Leader, and/or the Supervisor. Cases must be evaluated to:

- Eliminate the loss of potentially valuable information,
- Maximize the meaningful information obtained from the evidence,
- Determine if the requested examinations can be performed with the submitted evidence and with the available resources

Some of the considerations in evaluating the evidence include:

- The age of the evidence, especially when the evidence is biological material,
- The storage conditions of the samples prior to submission,
- Whether the liquid samples were dried before submission,
- Whether the evidence is moldy and/or putrefied,

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- Possible dilution of the samples
- Whether weapons or other objects require fingerprinting or have been fingerprinted,
- Whether all pertinent evidence has been requested,
- The availability of suspect known samples,
- The extent of screening required to obtain a search warrant for suspect known samples,
- The analyses that should be run if sample is limited,
- Possibility of sample remaining after analysis, and

- **Sampling and sample selection**

Sampling: Using part of a substance to represent the entire substance. The report will state conclusions about "the whole" based on testing a portion and there is the assumption of homogeneity (example, a portion of blood from a blood tube is collected and tested for DNA). Only reference samples will be considered homogeneous in the DNA laboratory. Saliva swabs, a portion of a FTA card, and a portion of liquid blood dried on swabs will be collected and will represent the whole. Only one extraction is needed for DNA (ISO 5.7.1, ISO 5.7.3). The report will indicate that "a swab" or "a portion" was used.

Sample selection: Selecting a sample of the whole based upon training, experience, competence, and the case scenario. Selection of samples for testing will be based on an attempt to determine which samples would yield the most probative information based on the case information. No assumption of homogeneity is made (example, testing 3 of many stains on a shirt for a presumptive test for blood).

- **Sample labeling (QAS 7.1.1c)**

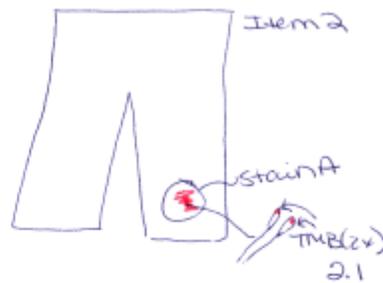
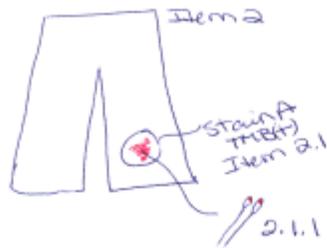
Each sample collected will be identified by the laboratory number and the unique LIMS number or sub-number for the case. The following guidelines also apply:

Serology: All positive stains (at a minimum) tested will be given a sub-item number and the report will specifically list which sub-items tested positive for the presumptive or confirmatory test and which ones were collected.

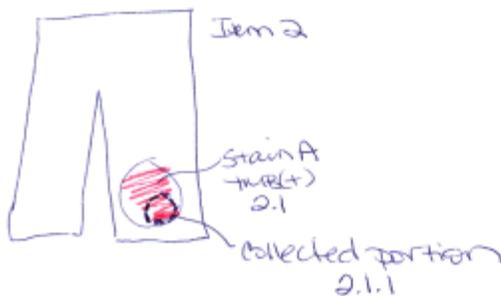
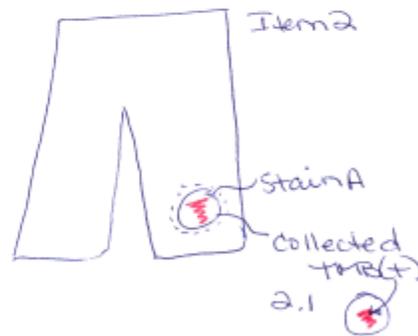
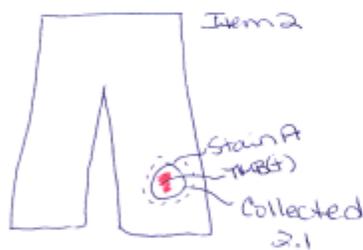
Below is an illustration of proper sub-item labeling.

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Swabbings



Cuttings



DNA: All samples tested for DNA will be identified by their sub-item number and will be reported as such. The samples selected for DNA will be based on analyst discretion and will serve the purpose of attempting to connect an evidence item with an individual.

- **Trace evidence**

Collection of trace evidence is at the analyst's discretion. If there is any concern trace evidence will be lost during analysis, it should be collected. If trace evidence is not going to be collected, extra caution should be used to ensure nothing is lost during analysis.

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➤ Hair

Occasionally, an investigation may be aided by the comparison of a questioned hair to known standards. DNA analysis may be performed on evidentiary hair only after:

- Complete information is derived from a microscopic examination of the hair by an approved laboratory, when applicable. For example, some cases with no suspects may require DNA testing without prior microscopic comparison. The following should be considered in evaluation of the case:

- What is the significance of the particular hair?
- Is it permissible (with the prosecutor or investigator) to destroy part of the evidence?
- Are there additional details of the case that may explain the hair?
- What is the condition of the hair, e.g., fragment, root, etc.? What is the likelihood of a DNA typing result?
- Is it desirable to postpone DNA typing at this time?
- Would mitochondrial DNA analysis by another laboratory be possible?

- **Chain of Custody (QAS 7.1.2)**

The LIMS system will be used to track the chain of custody of evidence within the Forensic Division with the exception of the packages containing the DNA cuttings and DNA extracts. These will be stored within the DNA lab freezer and will be tracked via paper chains (forms 17.36 and 17.37) stored in the case file. The time of creation of collected cuttings/swabs and DNA extracts (time at completion of extraction process) will also be documented on forms 17.36 and 17.37, as applicable. The DNA lab should maintain a photocopy or photograph of the APD paper chain of custody tags for the analyzed items in a case and these should be stored in the case record. Copies of the LIMS chains for parent items should be printed and placed in the case file. If an item is requested and possessed but not analyzed, a notation will be made somewhere in the case documentation (for example, on a worksheet or the copy of the chain of custody) and will also be mentioned in the report as receiving no analysis.

Bulk items of evidence will typically be received from the DNA CEL (central evidence locker). This is a secure locker area that only the DNA unit and property section have access to. For items received from the DNA CEL, the chain will have an entry showing a property section employee placing the item in the locker and the next entry will be when the DNA analyst retrieves the item from the locker. Once analysis is complete, the bulk items will be returned to the general return locker section of the CEL.

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- **DNA Analysis**

- **DNA Extractions**

The extraction of reference samples must be performed at a separate time or location from the extraction of evidentiary samples to eliminate the potential for reference to unknown sample contamination. It is also recommended that items of evidence from the suspect not be extracted adjacent to items of evidence from the victim.

- **Controls (QAS 9.5)**

A reagent blank will be extracted concurrently for each set of DNA extractions and will contain all reagents used in that extraction process (QAS 9.5.3.1). The reagent blank will be processed through the entire analysis, except quantitation, with the exception that it will not contain sample. If samples are concentrated on the speed vac the reagent blank will also be concentrated. A reagent blank must be analyzed with each PCR system (QAS 9.5.3.2a). Any remaining reagent blank should be stored frozen. The reagent blank shall be amplified on the same instrument model (QAS 9.5.3.2b) and in such a way that it will detect contamination in the most dilute evidence sample. For example, if 15 µl is the greatest amount of template amplified for any evidence sample in the batch, 15 µl of reagent blank must be used as template during the amplification (QAS 9.5.3.2c). The reagent blank will be run at the highest injection time of any of the samples within a case and on the same instrument model (QAS 9.5.3.3a, QAS 9.5.3.3b). It will also be analyzed at the lowest RFU value of the samples within a case (QAS 9.5.3.3c).

An amplification blank (negative control) will be introduced at the amplification setup step and will be included with each analysis step thereafter (QAS 9.5.2b). The amplification blank will contain all PCR setup reagents except DNA template. The amplification blank will be the last sample processed in the set and will be handled in such a way that it will detect contamination occurring during PCR setup. It will be amplified concurrently in the same instrument as the samples (QAS 9.5.2a).

An amplification positive control will also be introduced at the amplification setup step and will be included with each analysis step thereafter (QAS 9.5.2b, QAS 9.5.2b). It will be amplified concurrently with the samples in the same instrument and with the same loci the samples are analyzed with.

- **Consumption of evidence (QAS 7.2, QAS 7.3)**

The evidence quality and quantity will be preserved as much as possible without sacrificing the quality of the analyses. Whenever possible, at least half of the evidence sample will be preserved for possible re-analysis. Samples (i.e., cuttings or swabs) requiring depletion should have the substrate retained after extraction, when applicable. For questioned samples an approximate number of swabs (or the word "portion" for cuttings not depleted) of the sample used should be documented on the extract log. For reference samples, it will be assumed that at least half of the sample remains. If not, the amount used will be documented.

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➤ **Storage of evidence**

Biological evidence must be properly stored to preserve bio-chemicals assayed in body fluid identifications and DNA typing for current and future analyses. Storage conditions for all types of evidence present must be considered so that none are compromised.

Store sexual assault kits in the refrigerator or in a dry area at room temperature once received in the laboratory. Other items from a sexual assault case may be refrigerated as well depending on available space. Otherwise, store clothing, bedding, and other physical evidence in a dry area at room temperature until examined.

Blood cases containing small, dry items may be stored at room temperature. Refrigerate, do not freeze, liquid whole blood specimens until a sample is dried on FTA paper or swabs. Store larger items such as clothing, bedding, weapons, and other physical evidence containing bloodstains in a dry area at room temperature until examined.

After final analysis, store stains and extracts in the freezer. Repeated freezing and thawing of stains should be minimized. In the event that freezer space is exhausted, samples may be removed to a long-term evidence storage area (access limited to authorized lab personnel) for storage at room temperature. Casework cuttings/swabs and DNA extracts will be retained indefinitely as evidence. Post amplification product (work product) does not need to be retained (QAS 7.1.1b)

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Appendix 5A DNA Section Abbreviations

1, 2, 3, 4	AP strength of reaction weak to strong, e.g., 4+ for strong positive reaction
0	none
agg	aggravated
AE	additional evidence
ALS	alternate light source
AP	acid phosphatase
Apparent	app
CEL	central evidence locker
C/N	control negative
D	Depleted
dIH ₂ O	deionized water
Disp	Disposition
Epi, EC, EF	epithelial cell, epithelial fraction
Evid	evidence
F	frozen
F5	freezer 5
H	human
H/F	heads per field
K	known standard sample
L	Crime-Lite 2
LL	Crime-lite 82
M	manila
MM	master mix
ND	not done
NFR/PIC	Nuclear Fast Red/ Picroindigocarmine
NONA	not opened, not analyzed
NR	no reaction
NS	no stains seen
NSEV	no stains of apparent evidentiary value
NST, NSTE	no significant trace (evidence)
Obs	observed

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PB	presumptive blood
PCR	Polymerase chain reaction
PPF	PowerPlex Fusion
PT	purple top
PHT	phenolphthalein
Q	questioned sample
QP	quick prep
QNS	quantity not sufficient (for further analysis)
R/B	red/brown
Sus	suspect
S	stained
SA	sexual assault
S/I	sealed/initialed
S/I/D	sealed, initialed, dated
S/D	sealed, dated
SB	swab box
Sp	sperm
SP	swab packet
SF	sperm fraction
ST	Stochastic Threshold
Stn	stain
STR	short tandem repeat
TL	Tape lift
TMB	tetramethylbenzidine
TNTC	too numerous to count
U	unstained
UD	Undetected
Vag	Vaginal
V	victim
Vis	visible
w/	with
w	weak reaction

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Zip ziplock
[] concentrated
~ consistent with
() alleles below threshold

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5.2 Observation by Outside Experts

No Supplemental Requirements

5.3 Evidence Disposal

No Supplemental Requirements

5.4 Destruction of Hazardous Substances

No Supplemental Requirements

5.5 Outsourcing

No Supplemental Requirements

6 Laboratory Safety

No Supplemental Requirements

7 Personnel

7.1 Documents

No Supplemental Requirements

7.2 Subpoenas

No Supplemental Requirements

7.3 Private Case Consultations

No Supplemental Requirements

7.4 Testimony for Previous Employers

No Supplemental Requirements

7.5 Attendance

No Supplemental Requirements

7.6 Certification of Analysts

No Supplemental Requirements

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7.7 Employee Training Program (QAS 5.1.2)

The DNA section has a training manual that serves as an outline of the training program. Examiners will be provided fundamental serology (screening and body fluid identification) and DNA analysis training, which will include protocols in the SOP that are used in the lab. Serology and DNA training will be undertaken as separate units of training and will each conclude when:

- All competency samples are correctly analyzed;
- The competency notebook is approved by the Technical Leader;
- The competency notebook, other training records documenting completion of training requirements, and trainee credentials are reviewed by the Quality manager or Division manager;
- The trainee successfully completes a written exam administered by the Technical Leader.
- The laboratory supervisor and trainer(s), when applicable, recommend that the examiner be approved for independent casework;
- The Technical Leader recommends that the examiner be approved for independent casework; and
- The Quality Manager or Division Manager approves the examiner for independent casework.

Prior to completing DNA analysis training, examiners must additionally complete:

- Fifty (50) screening and/or body fluid identification cases.
- Six months of Forensic DNA analysis experience;
- One to five previously examined, adjudicated cases or mock cases, at least one of which includes a differential extraction of semen evidence, and the results of which must essentially agree with those of the qualified examiner; and
- The trainee will review 10 sets of data representative of casework and provide a written interpretation of the data according to the laboratory policy.
- Moot court to be evaluated by the trainer or the trainer's appointee.

In addition, each examiner's profile must be on file for the DNA typing systems in use in the laboratory. The DNA laboratory maintains a database of DNA profiles of personnel for the sole purposes of identification of the source of contamination and evaluation of in-house competency tests.

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Analysis training will be conducted according to the training manual. The Technical Leader has the authority to determine if some steps or processes in the training manual are not required for that particular trainee. In such cases, a memo will be included indicating the reason for the adjustments. Training beyond what is required in the training manual will be performed at the discretion of the Technical Leader and Supervisor.

The Technical Leader is responsible for proposing and reviewing changes to the training manual. The DNA Technical Leader is responsible for maintaining the training manuals, evaluating proposed changes and presenting those proposals for approval as appropriate. Experienced analysts new to the lab, at the discretion of the technical leader, may bypass the formal training program and immediately begin the competency test.

Competency Testing (QAS 5.1.2.2.1, QAS 5.1.2.2.3)

The trainee demonstrates competence in examinations through the preparation and presentation of a competency notebook of properly analyzed unknown (to the trainee) samples and the successful completion of a written examination. Previously approved analysts do not need to take the written test and can solely perform the practical test.

➤ **Competency notebook**

The DNA Technical Leader is responsible for creating, maintaining, and distributing training and competency samples as well as records of their sources and known types. The trainee will examine the competency samples according to the SOP, maintaining analysis documentation as for casework.

For serology analysis training, the competency samples will include a minimum of five stains and/or swabs for each of the training areas of blood and semen identification. For DNA analysis training, the competency samples will include a minimum of ten bloodstains, ten buccal swabs, five mixed stains of semen and epithelial cells for differential extraction, and five hairs (optional). The trainee may request hairs from up to eight sources and an unlimited number of hairs from any one source.

The Technical Leader will review the competency notebook prior to its presentation for review by the DNA Supervisor.

The competency notebook and/or training notebook will include at a minimum:

- The training log showing the start of training and the activities and date on each day of training;
- Analysis documentation for all competency samples; and
- Written tests taken during training.
- Other training records, such as practice sample analysis documentation, may be kept in the competency notebook.

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Written examination

The trainee must successfully complete a written examination covering the entire training unit of either serology or DNA prior to approval to perform independent analyses. The exam will be designed to document the trainee's knowledge of the procedures as well as the trainee's understanding of the theoretical basis for those procedures. In addition, the trainee may be asked to demonstrate knowledge and understanding of quality assurance practices relevant to his or her work. The exam will be administered by the DNA Technical Leader or designee.

Unusual samples

An approved examiner may use a valid procedure for analysis of a body fluid, tissue, bone or teeth not encountered during training providing the analyst has previously demonstrated competence in that procedure.

Experienced examiner competency testing of approved procedures (QAS 5.1.2.2.2)

Successful completion of a competency test is required prior to casework processing when an experienced DNA analyst begins employment at the APD DNA lab. An experienced examiner is required to test 5 previously analyzed samples in each method in order to demonstrate competency (**QAS 5.1.2.2.3**). Documentation will be retained in the laboratory. The technical leader will assess and document the adequacy of the analyst's previous training and a modified training program will be implemented for any required areas.

Experienced examiner competency testing for new procedures

Each experienced examiner will perform a minimum of 5 competency samples prior to using the new method in casework.

7.8 Employee Approval for Casework

No Supplemental Requirements

7.9 Employee Career Development

No Supplemental Requirements

7.10 Continuing Education (QAS 5.1.3)

Each examiner approved to perform DNA analyses will attend at least one continuing education training session (minimum of 8 hours) annually for the enhancement of DNA analysis skills. The DNA Technical Leader or Supervisor will recommend to management and coordinate training activities for personnel. Each examiner will place the certificate or agenda in their analyst notebook when he/she has completed any job-related training (**QAS 5.1.3.1.2**). For web-based continuing education, the training will be approved by the technical leader, the time of the training will be documented, and the completion of the training will be approved by the Technical Leader (**QAS 5.1.3.1.3**). The Technical Leader will have access to and ensure the maintenance of examiner training records in the laboratory.

Supplemental training will be used when remediation is needed. The Technical Leader will identify areas for which remediation is necessary based on the results of proficiency or

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competency test results, laboratory audits, or peer review activities and oversee or conduct such training using the training manual.

The DNA section will document the reading of articles by initialing the article as it is passed around. These articles will be retained in the Literature Review Binders (QAS 5.1.3.2).

7.11 Internship Program

No Supplemental Requirements

7.12 Volunteer Program

No Supplemental Requirements

7.13 Rider Program

No Supplemental Requirements